

# Evaluation of ethylene production by ten Mediterranean carnation cultivars and their response to ethylene exposure

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## Abstract

Vase life is one of the most important characters of postharvest cut flower quality. The onset of ethylene production and the amount of ethylene produced by flowers vary with the carnation cultivar, and thus influence their vase life. In the present study, differences in ethylene production and the response to exogenous ethylene among carnation cultivars were evaluated. Ten different cultivars: ‘Baltico’, ‘Domingo’, ‘Exotica’, ‘Famosa’, ‘Kiro’, ‘Madame Augier’, ‘Master’, ‘Mundo’, ‘Pilar’ and ‘Reina’ were studied. Five flowers for each cultivar were exposed for 8 hours to 1  $\mu\text{L L}^{-1}$  exogenous ethylene concentration. Ethylene production, fresh weight and water uptake was measured daily throughout the experiments. The 10 different cultivars studied showed clear differences in vase life, ethylene production, onset time in ethylene production and response to exogenous ethylene. The shortest vase life was for ‘Exotica’ flowers which was only 11.6 days, while ‘Baltico’ and ‘Pilar’ lasted 2.5-3 times longer than ‘Exotica’. Most of the investigated cultivars showed notable increases in the amount of ethylene. However, ‘Baltico’ and ‘Pilar’ flowers produced only a trace amount of ethylene and had the longest vase life. Results showed that cultivars with a long vase life (‘Baltico’ and ‘Pilar’) display high ethylene responsiveness and, in contrast, cultivars with a short vase life (‘Exotica’ and ‘Mundo’) present low responsiveness. The decline in fresh weight of cut flowers observed in the last phase of their vase life occurred earlier in short-lived cultivars than in the longer-lived ones.

**Additional key words:** exposure to ethylene; fresh weight; petal in-rolling; water uptake and wilting.

## Resumen

### Evaluación de la producción del etileno en 10 cultivares mediterráneos de clavel y su respuesta a la exposición al etileno

La vida útil de la flor cortada es uno de los caracteres más importantes de evaluación de la calidad en poscosecha. El momento y cantidad de etileno producido por la flor de clavel varía entre cultivares y eso influye en la vida útil de la flor. El objetivo de este trabajo fue evaluar diferencias en la producción de etileno y su repuesta al etileno exógeno en diez cultivares de clavel (‘Baltico’, ‘Domingo’, ‘Exotica’, ‘Famosa’, ‘Kiro’, ‘Madame Augier’, ‘Master’, ‘Mundo’, ‘Pilar’ y ‘Reina’). Se expusieron durante 8 h cinco flores de cada cultivar a una concentración de 1  $\mu\text{L L}^{-1}$  de etileno exógeno. Se midió la producción de etileno, peso fresco y absorción de agua diariamente. Los 10 cultivares estudiados presentaron diferencias claras en la vida útil de la flor, tiempo y producción de etileno y respuesta al etileno. ‘Exotica’ fue el cultivar con vida útil más corta (11,6 días), mientras ‘Baltico’ y ‘Pilar’ presentaron una vida útil 2,5-3 veces mayor que ‘Exotica’. La mayoría de los cultivares produjeron una notable cantidad de etileno, sin embargo ‘Baltico’ y ‘Pilar’, con la vida útil más larga, produjeron solo cantidades trazas de etileno. Estos cultivares de vida útil larga (‘Baltico’ y ‘Pilar’) mostraron alta sensibilidad al etileno, sin embargo, los de vida útil corta (‘Exotica’ y ‘Mundo’) presentaron baja sensibilidad. La reducción del peso fresco observado en la primera fase de la vida útil de la flor, ocurre antes en los cultivares con una vida útil corta que larga.

**Palabras clave adicionales:** absorción de agua; enrollamiento de los pétalos; exposición al etileno; marchitamiento; peso fresco.

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## Introduction

Prolonged vase life is one of the most important factors in terms of the quality of cut flowers. Thus, most horticulturists are involved to some extent in some aspects of postharvest horticulture, at least as consumers desiring ornamentals with an attractive appearance and long postproduction life (Kader, 2003). Strategies to prevent losses include the use of genotypes that have a longer postharvest life (Ebrahimzadeh *et al.*, 2008).

Carnation (*Dianthus caryophyllus* L.) is the main ornamental crop in Spain. Flower senescence in most carnation cultivars is characterized by autocatalytic ethylene production and subsequent wilting of the petals (climacteric flower) (Sato *et al.*, 2005), but some long lasting cultivar flowers are associated with reduction, delay or even the absence of ethylene (Wu *et al.*, 1991), thus influencing their vase life (Nukui *et al.*, 2004). The senescence of carnation flowers was first studied using the cultivar 'White Sim' (Whitehead *et al.*, 1984). In this cultivar, wilting of corolla is accompanied by a climacteric rise in ethylene synthesis (Fujino *et al.*, 1980) and a relatively short vase life (6-7 days). However, some later studies on other carnation cultivars such as 'Sandra' (Wu *et al.*, 1989), 'Killer' (Serrano *et al.*, 1991) and 'Master' (Serrano *et al.*, 1999) showed different behaviour and lasted much longer than 'White Sim'. They showed neither the normal increase in ethylene production, nor a marked respiration climacteric during their eventual senescence (Wu *et al.*, 1989), suggesting that the production of ethylene, or response to it, might vary among carnation cultivars.

The carnation flower is also highly sensitive to exogenous ethylene (Onozaki *et al.*, 2001) but the response to ethylene exposure also depends on cultivars. Some long-life cultivars, such as 'Chinera' and 'Epomeo', have lower ethylene sensitivities than that of the normal Sim-type cultivar 'White Sim' (Wu *et al.*, 1991). Onozaki *et al.* (2004) reported that Mediterranean carnation cultivars show more variation in ethylene production and sensitivity than standard Sim-type cultivars. Thus, it is important to know the ethylene production rate in commercial carnation cultivars and their response to ethylene.

In the present study, differences in ethylene production and their response to exogenous ethylene among carnation cultivars were evaluated.

## Material and methods

### Plant material

Ten carnation flowers of 10 different cultivars: 'Baltico', 'Domingo', 'Exotica', 'Famosa', 'Kiro', 'Madame Augier', 'Master', 'Mundo', 'Pilar' and 'Reina' were harvested from the Barberet & Blanc Company (Murcia/Spain) at the same development stage (when flowers were half-opened) and transported to the laboratory on the day of harvest. Stems were re-cut to 35 cm in length, placed in distilled water and held under controlled environmental conditions of 12 hours light, 12 hours darkness at  $21 \pm 1^\circ\text{C}$  and 60% relative humidity.

### Exposure to exogenous ethylene

For ethylene treatment, five freshly harvested flowers (one day after harvest) were enclosed in a 50 L glass chamber and exposed for 8 hours to  $1 \mu\text{L L}^{-1}$  ethylene (Sigma-Aldrich, Spain).

### Determination of vase life

The vase life of each flower was determined by the number of days after cutting until the petals showed in-rolling or browning and had no decorative value. Flowers were evaluated daily. Ethylene affected vase life (EAVL) was estimated as the ratio between the vase life of control flowers and the vase life of treated ones. Values of vase life were the means of 10 flowers.

### Measurement of ethylene production

Ethylene production was measured daily throughout the experiments. To measure the ethylene production, cut carnations were individually enclosed in 1 L glass jars for 1 hour. A 1-mL gas sample was then withdrawn and the ethylene concentration was determined by using a gas chromatograph (Varian 3900) fitted with a flame ionisation detector. Operating temperatures were: column  $80^\circ\text{C}$ , injector port  $150^\circ\text{C}$  and detector  $200^\circ\text{C}$ , using helium as a carrier gas at a flow rate of  $50 \text{ mL min}^{-1}$ . Ethylene production was expressed as nL of ethylene per gram fresh weight and hour.

## Fresh weight

Fresh weight (FW) of stems was measured daily with a Mettler Toledo balance with a precision of 0.001 g and was expressed as relative fresh weight (RFW) to the initial weight using the following formula:

$$\text{RFW (\%)} = (\text{FW}_t / \text{FW}_{t=0}) \times 100,$$

where  $\text{FW}_t$  is fresh weight of stems (g) at  $t$  days (2, 4, 6, etc.) and  $\text{FW}_{t=0}$  = weight of stems (g) at day 0. Relative fresh weight on day zero was established as 100%.

## Water uptake

Average daily water uptake was calculated from daily measurements of the weight of tubes without flowers as well as weight of water and flowers by using the following formula:

$$\text{WU} = (\text{W}_{(t-1)} - \text{W}_{(t)}) / \text{FW}_{t=0},$$

where WU = water uptake ( $\text{mL d}^{-1} \text{ g}^{-1}$  fresh weight),  $\text{W}_t$  = water weight (g) at  $t$  days (1, 2, 3, etc.),  $\text{W}_{t-1}$  = water weight (g) on the preceding day and  $\text{FW}_{t=0}$  = fresh weight of the stem (g) on day 0.

## Experimental design and statistical analysis

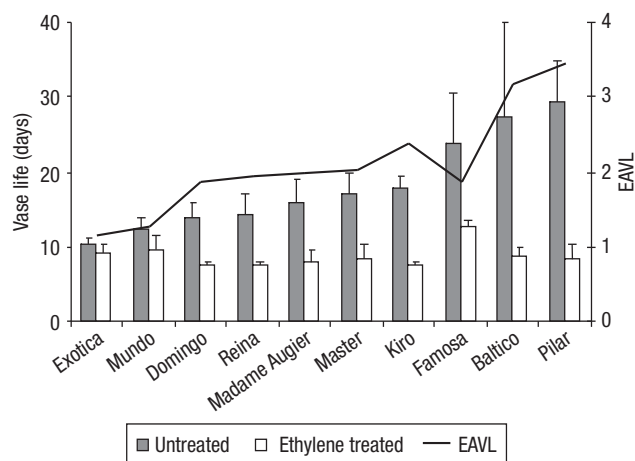
The experimental design was unifactorial (cultivar) with 10 repetitions per cultivar. An analysis of variance (ANOVA) and a least significant difference (LSD) by  $p < 0.05$  were conducted to determine the differences in ethylene production, vase life and percentage of fresh weight between cultivar.

## Results

### Vase life

Studied cultivars exhibited a wide range of variation in vase life (Fig. 1). 'Pilar' cut flowers had the longest vase life among the ten selected cultivars, followed by 'Baltico' and 'Famosa' flowers. These last two cultivars also showed a high standard deviation. The shortest vase life was for 'Exotica' flowers only being 11.6 days, while 'Baltico' and 'Pilar' lasted 2.5-3 times longer than 'Exotica' and 'Mundo' flowers.

Treatment of carnation cultivars with ethylene reduced postharvest life of all cultivars (Fig. 1). The

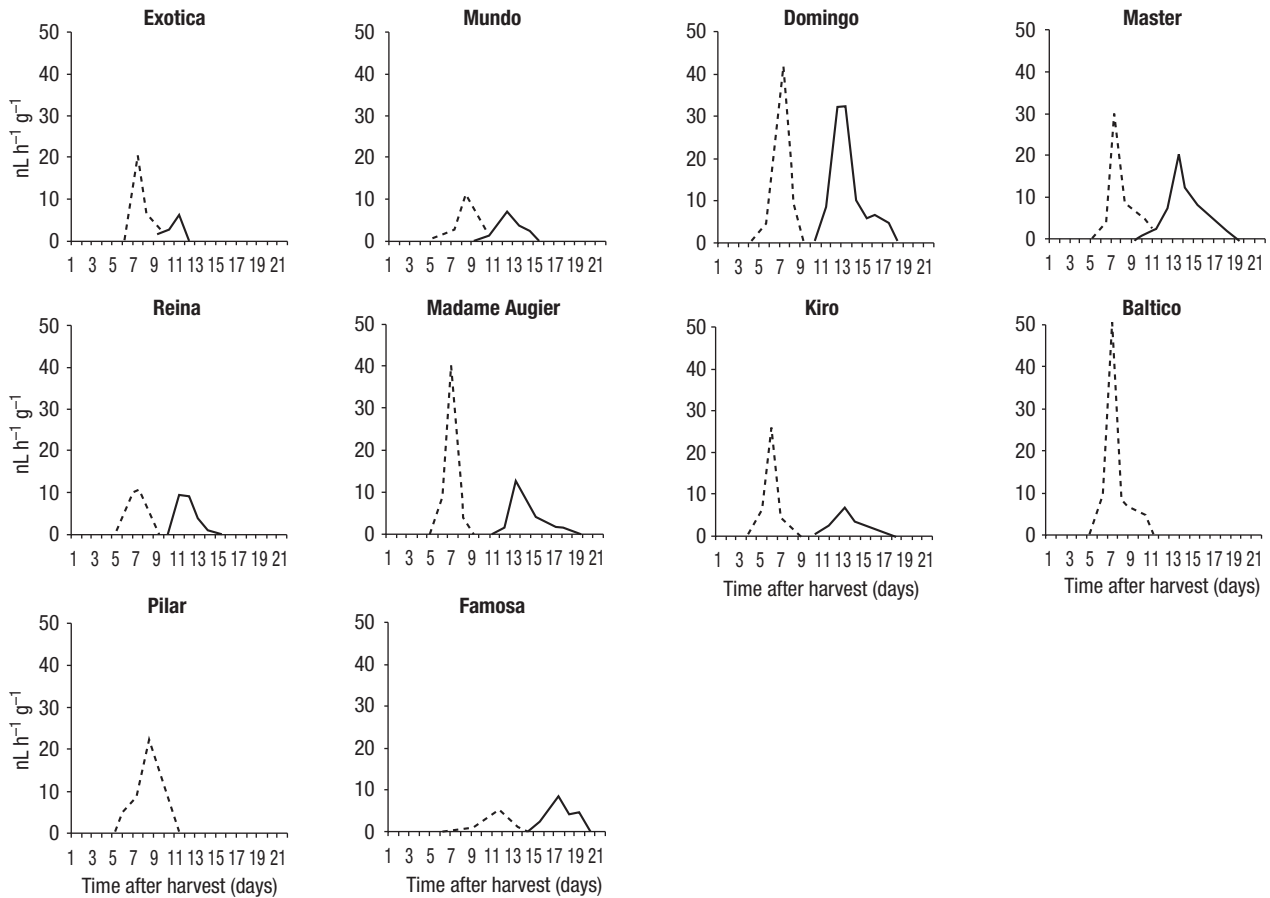


**Figure 1.** Vase life (VL) and ethylene affected vase life (EAVL) among ten treated with ethylene and untreated carnation cultivars. Values of vase life are the means of ten flowers ( $p < 0.05$ ).

mean vase life of the studied cultivars when exposed to exogenous ethylene was 7.5 to 10.0 days, whereas untreated flowers had more variant vase life in the wide range of longevity (11.6-30.5 days). On the other hand, the 'Famosa' cultivar had the longest vase life and the lowest ethylene production in response to exogenous ethylene. Ethylene affected vase life (EAVL) is presented in Figure 1. Cultivars were classified according to different values of EAVL: values around 1 as low response, values around 2 middle response and values near to 3 were classified as cultivars with high responsiveness. Results showed that cultivars with long vase life display a high EAVL ('Baltico' and 'Pilar'). It is assumed that the lack of ethylene production in these flowers (Fig. 2) resulted in a high EAVL (Fig. 1). Cultivars with mid-range vase life display a mid-level EAVL ('Domingo', 'Famosa', 'Kiro', 'Madame Augier', 'Master' and 'Reina') and cultivars with low longevity show a low EAVL ('Exotica' and 'Mundo').

### Ethylene production

In most cultivars, ethylene production was not detected during the first few days, but then most cultivars produced considerable amounts of ethylene. No ethylene was detected in the 'Baltico' and 'Pilar' cultivars until the end of their vase life (Fig. 2). Most of the investigated cultivars such as 'Domingo', 'Madame Augier' and 'Master' showed notable increases in the amount of ethylene and significantly decreased initiation time to produce ethylene. Moreover, results showed that



**Figure 2.** Ethylene production ( $\text{nL g}^{-1} \text{h}^{-1}$ ) in response to exogenous ethylene (---) compared with untreated (—) cut flowers in ten studied cultivars. Each time point represents the average value of ten flowers.

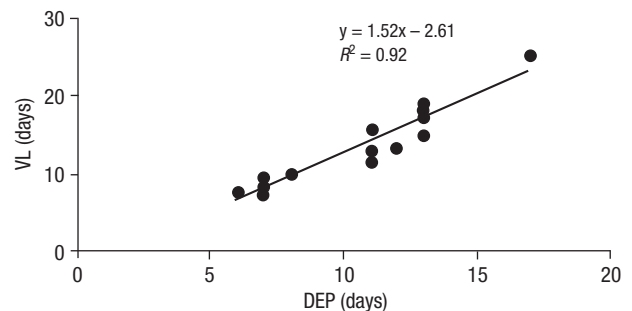
cultivars such as 'Baltico' and 'Pilar' that have no ethylene in standard atmospheric condition, showed significant amounts of ethylene after exposure to exogenous ethylene. On the other hand, 'Famosa' did not show any differences in the amount of ethylene between control and treated flowers, although ethylene production started earlier in treated flowers compared with untreated ones.

In addition, there was a correlation ( $R^2 = 0.92$ ) between days to ethylene peak (DEP) (*i.e.* days from harvest to reach maximum ethylene production) and vase life (VL) (Fig. 3).

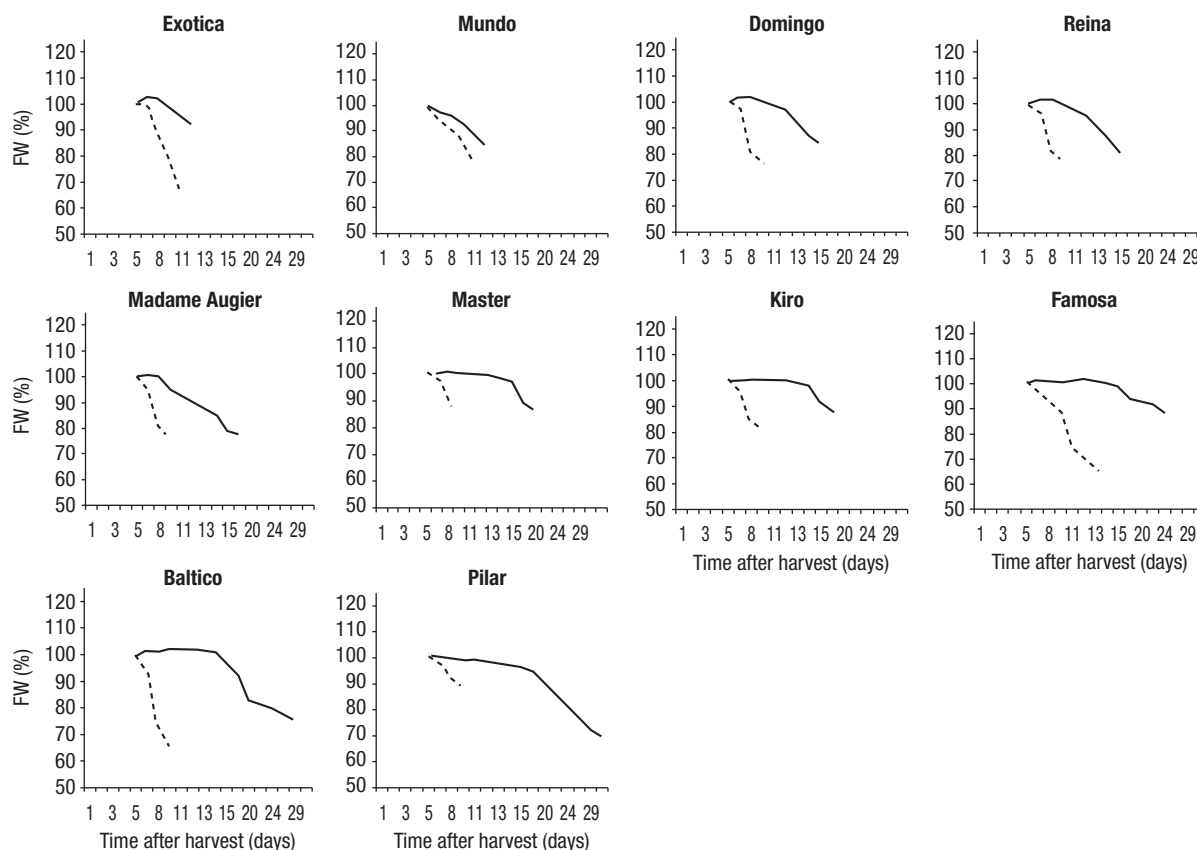
### Fresh weight

In the first few days, the fresh weight of most untreated flowers remained constant and after several days, RFW declined (Fig. 4). There were noticeable differences between cultivars. For example, 'Baltico', 'Famosa',

'Kiro', 'Master' and 'Pilar' maintained a higher relative fresh weight over a longer period, while other cultivars started to lose initial FW after only a few days. Relative FW loss was increased by the ethylene treatment and flowers showed apparent differences during the first



**Figure 3.** Relationship between days to ethylene peak (DEP) and vase life (VL) of 'Kiro', 'Madame Augier', 'Master', 'Famosa', 'Reina', 'Domingo', 'Mundo' and 'Exotica' cultivars of carnation treated and non-treated with ethylene. Values of vase life are the means of ten flowers.



**Figure 4.** Changes in fresh weight of cut flowers during postharvest life among studied ten carnation cultivars exposed (---) and not exposed (—) to ethylene. Each time point represents the average value of ten flowers.

few days. Among tested cultivars, ‘Mundo’ flowers were the least affected in fresh weight losses by exposure to exogenous ethylene.

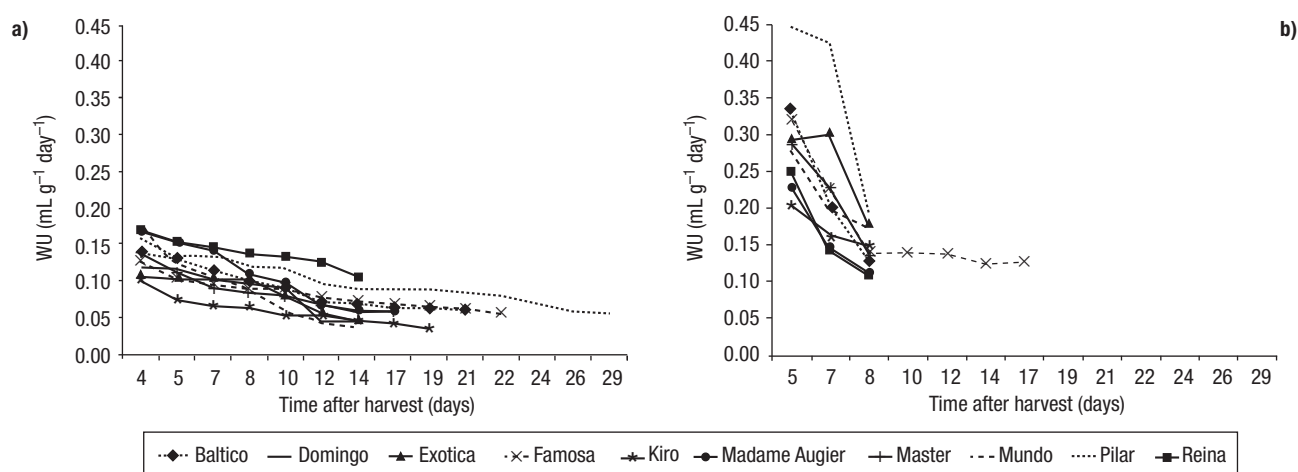
### Water uptake

Exposure of flowers to exogenous ethylene increased water uptake in cut stems, while untreated flowers in most investigated cultivars showed a slow decline in water uptake; flowers after exposition to ethylene declined dramatically water uptake (Fig. 5).

### Discussion

Most cultivated carnations behave as climacteric organs in which ethylene biosynthesis and respiration rate increase in association with the beginning of senescence (Van Altvorst and Bovy, 1995). Similar results have been found by Serrano *et al.* (1991) in the standard cultivar ‘Arthur’, by Wu *et al.* (1989) on

‘Sandra’ and by Peiser (1986) on ‘White Sim’ cultivars. In the present study, ethylene was not detected in ‘Baltico’ and ‘Pilar’ cultivars until the end of their long vase life, similar to that found by Serrano *et al.* (1999) on the ‘Killer’ cultivar. On the other hand, a good correlation between days after harvest to reached ethylene peak (DEP) and VL suggests that the system which postpones ethylene synthesis can increase vase life. Notable differences in ethylene sensitivity were observed among the ten investigated cultivars. Onozaki *et al.* (2008) showed that the variation in response time to ethylene treatment of the selected lines has a genetic cause. All evaluated cultivars in the present study were sensitive to exogenous ethylene, but large differences have been observed among them. The hypothesis that natural longevity was associated with low sensitivity to ethylene was not sustained (Müller *et al.*, 1998). Long-life cultivars such as ‘Baltico’ and ‘Pilar’ were very sensitive to exogenous ethylene, and in contrast, short-life cultivars such as ‘Exotica’ and ‘Mundo’ were less sensitive to exogenous ethylene. Thus, the results of the present study confirmed that longer vase life did



**Figure 5.** Differences in water uptake (WU) among tested cultivars of carnation not exposed (a) and exposed (b) to exogenous ethylene. Values of vase life are the means of ten flowers ( $p < 0.05$ ).

not result from lower ethylene sensitivity, but rather from lower ethylene production. Similar results have been reported by Onozaki *et al.* (2006) on carnation cultivars and selected second- and third-generation lines.

The decline of fresh weight is due to transpiration exceeding water uptake (Harlevy and Mayak, 1981). One of the factors that influences the water deficit is the rate of water uptake and transport as well the ability of different organs on the cut flower shoot to compete for water which may be in limited supply (Halevy and Mayak, 1981). Ethylene treatment apparently changed the pattern of relative fresh weight loss. Exogenous ethylene accelerates the senescence through the stimulation of ethylene biosynthesis associated with an increased respiration rate and consequent increases in fresh weight losses (Ebrahimzadeh *et al.*, 2008).

Senescence of most carnation cultivars in the present study is characterized by petal in-rolling and wilting which agrees with the results obtained by Nukui *et al.* (2004). However, 'Baltico' and 'Pilar' cultivars showed petal colour fading and browning at the end of their vase life.

Exposure to exogenous ethylene promotes senescence of climacteric flower petals with activation of 1-aminocyclopropane-1-carboxylic acid (ACC) syntheses and/or ACC oxidase (Borochoy and Woodson, 1989) and reduces vase life. It has been reported that the sensitivity to ethylene is correlated with vase life in carnation flowers that significantly varied among cultivars (Onozaki *et al.*, 2001, 2008). These findings agree with earlier studies by Serek *et al.* (1994) on *Rosa hybrida* 'Victory Parade'.

As final conclusions, treatment of cut carnation cultivars by exogenous ethylene reduced postharvest

life in most of the cultivars with an early start in ethylene production and synthesis rate, compared with untreated flowers. Nevertheless, notable differences in ethylene sensitivity were observed among the ten investigated cultivars. Results showed that cultivars with high vase life display high ethylene responsiveness and in contrast cultivars with low vase life display low responsiveness. The time to reach maximum ethylene production was highly correlated with their vase life. The decline in fresh weight observed in cut flowers occurred earlier in short-life cultivars than in the longer life ones, and ethylene treatment apparently changed the pattern of relative fresh weight loss. However, water uptake is higher in those flowers exposed to exogenous ethylene.

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